

Claims

1. A method for producing a polypeptide product which is substantially free of an undesired protein, the process comprising culturing a host cell which is able to express said polypeptide product and which is able to express said undesired protein only in a mutant form which form has the activity of the corresponding native protein under culture conditions but is unstable under conditions at which the said polypeptide product remains stable; and recovering the desired product, wherein either the host cell culture or the recovered product is subjected for a sufficient period of time to conditions under which the undesired protein is unstable so as to denature the undesired protein.

2. A method according to claim 1 wherein the host cells are cultured for a period which is sufficient to allow production of polypeptide product, and then a batch of said culture is subjected to the said conditions under which the undesired protein is unstable for a sufficient period of time to denature the undesired protein, and the polypeptide product is recovered.

3. A method according to claim 1 or claim 2 wherein the conditions at which the undesired protein is denatured and the polypeptide product remains intact are temperature conditions.

4. A method according to claim 3 wherein said temperature conditions are elevated temperatures.

5. A method according to claim 4 wherein the elevated temperature is 37°C or more.

6. A method according to claim 1 or claim 2 wherein the conditions at which the undesired protein is denatured and the polypeptide product remains intact are pH conditions.

7. A method according to ~~any one of the preceding claims~~ wherein the desired polypeptide product is luciferase and the undesired protein is adenylate kinase.
8. A method according to claim 7 wherein the adenylate kinase is thermolabile at a temperature of 37°C or more.
9. A method according to claim 7 ~~or claim 8~~ wherein the adenylate kinase includes mutations at amino acids 87 or 107 in the sequence of the *E. coli* adenylate kinase.
10. A recombinant cell which comprises a first nucleotide sequence which encodes a desired polypeptide under the control of regulatory elements which allow expression of said polypeptide, and wherein ~~a gene which encodes a protein which is undesirable as a contaminant in preparations of said polypeptide product is mutated such that the protein expressed is unstable under conditions in which the polypeptide product remains stable.~~
11. A recombinant cell according to claim 10 wherein the said desired polypeptide comprises a luciferase and the said undesired protein comprises adenylate kinase.
12. A recombinant cell according to claim 10 ~~of claim 11~~ which further comprises at least one selection marker.
13. A recombinant cell according to ~~any one of claims~~ 10 to 12 which comprises a prokaryotic cell.
14. A recombinant cell according to claim 13 which comprises a recombinant *E. coli* cell.
15. A method for producing a recombinant cell according to ~~any one of claims~~ 10 to 14 which method comprises in any order (a) transforming a host cell with a vector which encodes said undesired protein in a form which is unstable under given conditions, subjecting transformants to said conditions and detecting those in which protein product is denatured, and (b) transforming said host cell with a vector which encodes a desired polypeptide which is

stable under said conditions and a first selection marker, and using the first selection marker to detect stable transformants.

16. A method according to claim 15 wherein the vector which encodes said undesired protein in a form which is unstable under given conditions further comprises a selection marker which is different to said first selection marker, and stable transformants are selected.

17. A method according to claim 15 ~~or claim 16~~ wherein said selection markers comprise particular different antibiotic resistance genes.

18. A polypeptide product which is substantially free of an undesired protein, as obtained by a method according to ~~any one of claims 1 to 9~~.

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